ARMY

Submission of Proposals

The responsibility for the implementation, administration, and management of the U.S. Army Small Business Technology Transfer (STTR) Program rests with the Army STTR Program Manager at the U.S. Army Research Office (ARO). You are invited to submit STTR proposals to ARO at the US Postal or physical addresses below. Proposals must be received at ARO no later than the required solicitation closing date and hour.

Physical Address for Private Delivery Services
U.S. Army Research Office
ATTN: STTR-99 (LTC Jones)
4300 South Miami Blvd
Research Triangle Park, NC 27703-9142
Telephone: 919-549-4200

Mailing Address for U.S. Postal Service
U.S. Army Research Office
ATTN: STTR-99 (LTC Jones)
P.O. Box 12211.
Research Triangle Park, NC 27709-2211

The Army has identified nine topics, numbered ARMY 99T001 through ARMY 99T009, to which small businesses and their partner research institutes may respond. Only proposals addressing these topics will be accepted for consideration for Phase I of the Army STTR Program.

The Army anticipates sufficient funding to allow award of one to three STTR Phase I contracts to firms submitting the highest quality proposals in each topic area. Awards will be made on the basis of technical evaluations using the criteria contained in the solicitation within the bounds of STTR funds available to the Army at the time of award. If no proposals in a topic merit award relative to the proposals received in other topics, the Army will not award any contracts for that topic.

Proposals for Phase I are limited to a maximum of $100,00 over a period not to exceed six months.

Any Phase II contracts resulting from Phase I proposals submitted for this solicitation will be limited to a maximum of $500,000 over a period of two years. Phase II contracts will be structured as a single year contract with a one year option.

Army STTR FY99 Topic Descriptions

ARMY99T001 TITLE: Hybrid Optical/Digital Imaging

TECHNOLOGY AREAS: Sensors

OBJECTIVE: Develop hybrid optical/digital imaging systems which have enhanced performance compared to systems using conventional optical components.

DESCRIPTION: For many visual and infrared imaging systems, as well as other optical and electro-optical systems, the lenses and optical components represent a significant portion of the total system cost. This cost penalty is not only in terms of dollars, but also involves other parameters such as size and weight. Recent research has demonstrated the potential for enhanced performance with optical systems in which the wavefront image is initially encoded by tailored phase masks, and subsequently decoded with digital post-processing of the detected image. With appropriately designed masks, it is possible to reduce a number of aberrations including chromatic [1], spherical, and aperture limited depth of focus [2]. These sorts of enhancements have the potential to provide a wide range of significant system improvements. For example, increased depth of focus could allow the production of high fidelity, focus-free video cameras, and increased tolerance to field curvature could allow the design of improved flat-bed scanners. Also, these performance enhancements could be traded-off against other constraints in the overall system design. For instance, the performance of inexpensive plastic lenses might be improved to the point that they are viable for demanding applications; and potential decreases in size and weight, while maintaining or improving performance, are critical considerations for DoD systems. Successful commercialization of this technology will require a better understanding of the design of the phase masks, development of optimized post-processing algorithms, and an improved understanding of the implications for overall system design.

PHASE I: Develop techniques for the design and analysis of hybrid digital/optical systems. This should include techniques for developing the appropriate phase masks for the desired response, and optimization of the digital signal processing for near real-time speed and ease of implementation.

PHASE II: Apply the results and techniques developed in Phase I to the design and analysis of prototype unconventional optical components, including a cost/benefit analysis for incorporating the components in various systems.

PHASE III DUAL USE APPLICATIONS: Hybrid digital/optical imaging has the potential to significantly impact numerous DoD and commercial markets. Large performance gains and/or cost reductions can be expected for equipment ranging from the massive video camera market to the numerous military optical and electro-optical systems.

KEY WORDS: hybrid optical/digital, optical aberration, wavefront encoding.

REFERENCES: 1. H. B. Wach, W. T. Cathey, and E. R. Dowski, Jr., "Control of chromatic focal shift th rough wavefront coding", to be published in Applied Optics, August, 1999. 2. E. R. Dowski, Jr. and W. T. Cathey, "Extended depth of field through wavefront coding", Applied Optics, 34, 1859 (1995).

ARMY99T002 TITLE: Activable Molecular Strucure Probes for Biological Detection

TECHNOLOGY AREAS: Chemical/Bio Defense

OBJECTIVE: Characterize, for purposes of manipulation and exploitation as unique products in state-of-the-art biological detection strategies, biomolecular or biomimetic components able to function as high specificity, high sensitivity recognition and signalling elements for biological threat agents, as represented by any member of that general class, widely defined.

DESCRIPTION: Advances in fluorescence probe and other reporter molecule technology are emerging rapidly in biological sciences applications, as are some far-reaching possibilities for application offered by innovations in optoelectronic materials and nanotechnology. Along with parallel technological breakthroughs in the imaging sciences and in our understanding of the conformational events comprising biomolecular signal transduction, these advances should allow major leaps forward to enhance our capabilities for point and remote detection of biological threat agents of concern to counter-threat and counter-proliferation operations.

PHASE I: Provide feasibility of approach, in results of initial experimental and conceptual studies, for meeting one or more of the criteria described here as areas of interest. a) Establishing Specificity for Targeting: Synthesis and characterization of ligands able to recognize and bind highly selectively, in the presence of many competing molecules, to target biological macromolecules such as proteins, nucleic acids, or other components of threat agents of biological origin. b) Signal Emitters: Innovative biochemical and biophysical approaches toward generation of an optical, electronic or mechanical signal suitable for reception and further processing such as amplification with high signal to noise properties built in. c) Signal Production/Transduction: Generally applicable structural basis for potential signalling pathways using such concepts as induced conformational change, relying on the ability of threat agents of biological origin to promote structural reorganization in or around the cellular target biological receptor with which they interact to cause their toxic effect.

PHASE II: Implementation of research results from Phase I in high-selectivity recognition and signalling component(s) with demonstrable potential for incorporation as functional "sense and respond" material for biological detection system. Establish foundation for eventual biological or biomimetic molecular manufacturing of prototype biological detection system component. Offerors are encouraged to explore issues of compatibility with ongoing systems modernization efforts in the DoD Chemical and Biological Defense Program, but in any case, research and development approaches to be transferred must be entirely unique and innovative, and not simply extensions of existing technology.

PHASE III DUAL USE APPLICATIONS: Production of items of interest to the medical, agricultural and environmental sciences communities, including, but not limited to (1) medical diagnostics for pathogen and biological toxin detection and identification, (2) ensuring food and crop quality and monitoring field application of genetic manipulation for improvement in crop yield and pest resistance management, (3) process monitoring in biotechnological manufacturing, and (4) monitoring of native and engineered microorganism field status for environmental bioremediation.

KEY WORDS: biological detection, biomolecular probes, signal transduction, conformational change, reporter molecules

REFERENCES:

1. Farrens,D.L., Altenbach,C., Yang, K., Hubbell, W.L., & Khorana, H.G. (1996). Requirement of rigid-body motion of transmembrane helices for light activation of rhodopsin. Science, 274, 768-770.
2. Braha, O., Walker, B.,Cheley, S., Kasianowicz, J.J., Song, L., Gouaux, J. E., & Bayley, H. (1997). Designed protein pores as components for biosensors. Chemistry and Biology, 4, 497-505.
3. Hellinga,H.W. & Marvin, J.S. (1998). Protein engineering and the development of generic biosensors. Trends in Biotechnology, 16, 183-189.

ARMY99T003 TITLE: The Microbial Ecology of Contaminant Destruction

TECHNOLOGY AREAS: Environmental

OBJECTIVE: To optimize the most promising approaches towards exploitation of microbial systems for economically and environmentally sound contaminant destruction.

DESCRIPTION: The DOD is faced with a variety of problems related to soil, sediment, and ground water contamination. The explosives TNT, RDX, and HMX are of particular concern because former manufacturing, handling, and storage activities have resulted in contaminated soils where explosives and /or their toxic residues are leaching into ground water supplies. Numerous military installations and defense manufacturing facilities also

have spill sites contaminated with Petroleum, Oil, & Lubricants (POLs), Polychlorinated Biphenyls (PCBs), heavy Polynuclear Aeromatic Hydrocarbons (PAHs), solvents and lubricants. The DOD is committed to a policy of environmental stewardship, one pillar of which is the clean-up of contaminated defense sites. Presently, more than 10,000 defense sites require the clean-up of organic-based contamination at a cost in excess of $20B. Site managers are faced with a complex problem characterized by heterogeneous soils and biogeochemical environments with mixtures of contaminants and their decomposition products. Presently, the only treatment for these soils is incineration, which is expensive and politically problematic. Proposals should address the biochemical and physiological mechanisms underlying biodegradative processes, as well as specific organisms and consortia of organisms to attain complete mineralization of the organic contaminants. Other areas to consider include biodegradation kinetics (rate limiting steps), aerobic versus anaerobic systems, the effects of mixed contaminants, the fate of metabolites and the fate of any added organisms. The interaction between various types of soil and contaminants or microbes is important for bioavailability concerns.

PHASE I: The initial focus of the work will be on developing the basic biochemical, microbial, and genetic data for bioremediation of military-unique energetic compounds such as TNT, RCX and HMX. The effort will be directed toward degradative pathways, complexities of inter-microbial interactions, and the interactions of microbes with both contaminants and the components of the natural environment at the nanometer to pore scales. The issues of biosurfactants (natural or added), bioavailability, and genetic engineering, are also of interest.

PHASE II: The Phase II effort will be directed toward the scale-up of the most promising results from Phase 1 through proof-of-principle demonstrations at the pilot and field scale, with the ultimate aim of improving on those results in a manner that would be cost-effective for application to the clean-up of contaminated sites on military installations.

ARMY99T004 TITLE: Robust Weather Radar Algorithms for Hydrometeorlogical Forecasting and Analysis

TECHNOLOGY AREAS: Battlespace

OBJECTIVE: To produce and validate robust radar-rainfall algorithms, innovative hydrometeorological data assimilation procedures, and physics-based distributed hydrologic models incorporating uncertainty for emerging weather radar products generated under the joint DOD-NOAA-FAA NEXRAD program.

DESCRIPTION: The recently commissioned national network of WSR-88D (Weather Surveillance Radar, 1988 Doppler) weather radars, also known as the NEXRAD system (Crum et al., 1993) deployed and operated by DOD, NOAA, and FAA provides unprecedented fine space-time scale resolution observations of precipitation. The spatially-lumped hydrologic models in widespread use today cannot take full advantage of the space-time precipitation estimates provided by the WSR-88D weather radar system. However, such observations are being utilized in a new generation of distributed hydrologic models that are capable of emulating the influence of spatio-temporal patterns of precipitation and complex topography on hydrologic response. Currently, there is an emerging R&D effort in the area of weather-related services by universities and private industry that is directed toward the provision of user-specific hydrometeorological forecasting. New distributed hydrologic models that use the full information content of WSR-88D precipitation estimates and distributed land-surface topographic data have the potential to be commercially valuable to the hydrologic engineering community. This particular topic offers an opportunity to rapidly bring this promising area to full maturity and integrate: (i) government implementation of advanced earth-observing systems, (ii) basic research, and (iii) private-sector activities to meet critical and emerging needs in the area of real-time hydrologic forecasting/analysis and water resources management.

The modern WSR-88D weather radar network produces massive quantities of data about the occurrence of storm precipitation. However, conversion of WSR-88D observations to precipitation estimates is a very complex and difficult task (Zawadzki, 1984; Smith et al., 1996). The following needs must be satisfied to realize the full hydrologic forecasting potential of this information: (i) development and testing of robust radar-rainfall estimation algorithms for different hydroclimatological regimes, based upon NEXRAD weather radar products as delivered; (ii) creation of new net-aware software tools for efficiently storing, managing, and processing the tremendous quantities of data produced by the WSR-88D network; (iii) new and innovative hydrometeorological data assimilation procedures; and (iv) refinement of physics-based, distributed hydrologic models that are capable of

real-time integration of observations from multiple sensors/sources and optimized to efficiently utilize the full information content of WSR-88D precipitation estimates and fine spatial resolution land-surface characteristics data in a manner that considers the influence of uncertainty. Of particular interest are algorithms that address advection, raindrop diameter distribution variability, orographic and range effects, and evaporation. This development should be performed within the context of the Army WMS/CASC2D hydrologic modeling approaches. A critical issue to be addressed is how to assimilate large volumes of overlapping data with differing uncertainty structures into a consistent set of algorithms for quantitative, space-time precipitation estimation. Such space-time precipitation estimates will be used in the context of physics-based rainfall-runoff models for soil moisture accounting and flood prediction. Modern weather radar approaches to precipitation estimation and a new capability for hydrologic forecasting and analysis that utilizes this information has the potential to provide significant improvements in both CONUS military mobility modeling and river stage forecasting, as well as in the planning and execution of CONUS training exercises. Longer lead time forecasts and advanced warning of unusual hydrologic conditions will also benefit the water resources management, reservoir control, and river navigation management missions of the Corps of Engineers through flood hazard mitigation. The development of continuous soil moisture accounting over complex topography may also provide the initialization field for numerical weather prediction models and mobility/trafficability products for military vehicles, together with validation data for retrieval algorithms for soil moisture estimation based on emerging remote sensing approaches. The extension of such modern tools and techniques over the vast domain of the NEXRAD system coverage will also permit the quantitative evaluation of scaling relationships for hydrologic fluxes and soil moisture from hillslope, to watershed, to regional scale. The design of military and civilian engineering structures, as well as environmental management of military and civilian land, will benefit significantly from integrated hydrologic and hydraulic modeling systems that utilize radar rainfall products.

PHASE I: Initial activity will involve technological research leading to (i) the operational development and testing of robust radar algorithms which are generally applicable to different types of precipitation, the specifics of which will be defined through consultation with USAE Waterways Experiment Station (WES), and (ii) rigorous network-wide validation procedures for calibration and verification of radar-based rainfall estimates against surface instruments.

PHASE II: The follow-on effort will develop novel hydrologic-hydraulics modeling approaches which fully utilize the NEXRAD space-time precipitation estimates and deliver to the Army a the fully- documented and validated weather radar algorithms, data assimilation procedures, and hydrologic models generated during the Phase I and II efforts.

PHASE III DUAL USE APPLIACTIONS: The technology developed under this topic would have broad applicability to the military, commercial, and private sectors. The estimated space-time rain-rate fields will be utilized by hydrologic models for complex terrain to generate soil moisture estimates, streamflow forecasts, and engineering design applications.

KEYWORDS: NEXRAD, weather radar, precipitation monitoring, hydrometeorological analysis and forecasting, hydrologic modeling

REFERENCES:

1. Crum, T.D., R.L. Alberty, and D.W. Burgess, 1993, Recording, Archiving, and Using WSR-88D Data: Bull. Amer. Meteor. Soc., 74:645-652.

2. Smith, J.A., D.J.Seo, M.L. Baeck, and M.D. Hudlow, 1996, An Intercomparison Study of NEXRAD Precipitation Estimates: Water Resources Research, 32:2035-2045

3. Zawadzki, I., 1984, Factors Affecting the Precision of Radar Measurements of Rain: Proc. 22nd Conference on Radar Meteorology, Amer. Meteor. Soc.: 251-256.

ARMY99T005 TITLE: Next Generation Long-Term Electroencephalogram Recording Devices for Mice

TECHNOLOGY AREAS: Biomedical

OBJECTIVE: To develop a cost-effective monitoring system capable of long-term EEG recordings in mice to facilitate basic molecular and genetic sleep research.

DESCRIPTION: The mouse is an important model system for studying sleep, as well as sleep disorders, sleep deprivation, and control of the sleep cycle. Stable, long-term recordings of the electroencephalogram (EEG) from the mouse brain are essential for determining the sleep-wake behavioral state of the animal. The current technology is dependent on recordings generated from a tethered mouse with a surgically implanted swivel, connected to the recording device. This system significantly encumbers the mouse and may affect its behavior. Also, because of the surgery required and the limitations of this recording device, it is impossible to follow the behavior of large numbers of animals. A technological advance facilitating the screening and monitoring of many animals would enable researchers to take advantage of recent progress in molecular and genetic sleep research. One possibility is to develop a telemetry-based recording system. However, there are several technical issues that must be reconciled. The first is that the weight of the telemeter unit should not exceed 500mg, and it should be no larger than 4mm x 4mm x 2mm. It must also be able to function regardless of the animal's position in the cage, and the power requirements must be met when the animal inclines its head up to 45 degrees of horizontal. Also, the power unit should fit the top of a standard mouse cage (7" x 10") and should not interfere with intake of food or water. Ideally, one power unit could serve several cages. Another issue to consider is the possibility of signal interference if the researcher is simultaneously recording the behavior of several animals.

PHASE I: Development of a prototype long-term EEG monitoring system, in conjunction with sleep researchers, for the mouse.

PHASE II: Refinement of the phase I effort to resolve any technical difficulties and reduce costs as necessary. Comparisons should be conducted with existing technologies with particular attention paid to the qualitative and quantitative similarities and differences in the data sets obtained.

PHASE III DUAL USE APPLICATIONS: Sleep disruption and sleep disorders are significant problems in both civilian and military communities. Shift workers, emergency personnel, health professionals, and soldiers alike will benefit from molecular and genetic understandings of sleep need and vulnerability to sleep loss. Much of this work will be initially conducted in animal models, such as the mouse, and a less cumbersome, high throughput, cost effective monitoring system is essential to be able to capitalize on current advances in sleep research.

KEY WORDS: electroencephalogram, EEG, telemetry, sleep, mouse

ARMY99T006 TITLE: Creating Knowledge and Improving Training Through Latent Semantic Indexing

TECHNOLOGY AREAS: Manpower

OBJECTIVE: The objective is to extend and exploit current statistical models of human cognition, learning, and understanding to create new textual exercises, plans, summaries, testing and training materials from text knowledge bases of logistic command post exercises, ARTEPS, field manuals, and written military testing and training materials.

DESCRIPTION: A major requirement of all good staff work is the creation of multiple plans that implement the commander's intent and prepare for all major contingencies with risky but aggressive routes of attack. During real battle and training exercises there are never sufficient alternatives available, nor are these updated to reflect ongoing changes; nor are there automated means available to support staff tasks in a flexible and creative way. Recent innovations in statistical analysis of text for the enormous resources on the internet, such as Latent Semantic Analysis (LSA), have created proven tools that can index text, and judge its semantic relationships in ways that fundamentally mimic human knowledge representation and learning. This research and development

will extend and modify those tools so that they actually produce in an automated way, new creative exercises, plans, summaries, testing and training materials from existing textual databases.

PHASE I: Based on a thorough understanding of the empirical and theoretical literature on human knowledge representation and learning, develop a mathematically and psychologically sound approach, such as one based on LSA, to automatically generating new plans and exercises from existing text databases. The conceptual framework must explicate the choice of information theoretic, statistical, and multivariate approaches to adapting LSA to this novel purpose of generating new, creative, and semantically coherent plans and exercises.

PHASE II: Exploit the framework developed in Phase I to create useful plans in potential or actual logistic exercises. Assess and evaluate the exercise and plan's strengths and shortcomings, and reiterate the development process to improve the training effectiveness by addressing identified issues.

PHASE III DUAL USE COMMERCIALIZATION: Apply the technology for interpreting massive text databases in applications selected from many potential sectors, such as (for example): to generate summaries and meaningful descriptions of internet sites for search engines; to create meaningful summaries and educational materials for high school and college level technical courses; or to develop automated tools for developing plans and reports in many commercial enterprises.

KEY WORDS: Command and Control, Logistics, Staff Planning, Latent Semantic Analysis, Training, Human Knowledge Representation.

REFERENCES: Landauer, T. K, & Dumais, S. T. (1997) A solution to Plato's Problem: The Latent Semantic Analysis theory of acquisition, inductions, and representation of knowledge. Psychological Review, 104, 211-240.

ARMY99T007 TITLE: Wastewater Treatment Technology for Temporary Base Camps

TECHNOLOGY AREAS: Environmental

OBJECTIVE: To develop a filtration unit for removal of particulate matter from wastewater generated at temporary base camps. This unit will be used in place of large settling tanks which are cumbersome to deploy and produce a contaminated sludge that must be dewatered and disposed of by soldiers or contract operators.

DESCRIPTION: The Army has identified a need for a deployable wastewater treatment system for use at temporary base camps such as those in Bosnia. Conventional package plants are not suitable for this application because they were designed for permanent installations like suburban subdivisions and mobile home parks. They are heavy, expensive, and not configured for deployment. Also, they produce a sludge which must be properly disposed of to prevent it from becoming a health hazard to soldiers and civilians in the theater of operations.

Instead of settling out the particulate matter in wastewater, it is possible to filter it out with a cheap filter that is subsequently incinerated. This would eliminate the large settling tanks and the sludge disposal problem. Previous studies conducted at CRREL (Martel et al., 1999), indicate that nonwoven geotextiles can remove 70% of the total suspended solids (TSS) and 40% of the biochemical oxygen demand (BOD) from raw wastewater, which is equivalent to primary treatment. Other filter media such as recycled paper may also be feasible.

The proposed wastewater filtration unit (WFU) would be a standalone unit that could both filter the wastewater and incinerate the filter. The unit should be enclosed to keep out flies and other vectors, and to control odors. From an operational viewpoint, it would be advantageous to have automatic advancement of the filter medium as it becomes clogged. The clogged portion would then be incinerated. The only byproduct of the WFU would be a small amount of ash that would be collected and disposed of in a safe manner. The WFU could be used at several locations in the waste stream. It could be used to remove food wastes and other solids from a graywater line. It could be used as a primary treatment process prior to a secondary treatment process such as a trickling filter and it could be used to filter the effluent from a trickling filter. Performance characteristics and operational variables are expected to be different for each application. However, from a logistical point of view it would be advantageous to have a single unit that can accommodate a wide range of applications and flows. Large flows could be handled by connecting several units in parallel.

PHASE I: Proof of concept analysis including bench scale studies, preliminary design, deployability evaluation, operation and maintenance assessment, regulatory requirements, and cost estimate.

PHASE II: Demonstration of technology in a pilot or full scale unit configured for deployment, including the development of operational information on treatment efficiency and hydraulic capacity of the filter, incineration of filter media, odor control, and ash characteristics.

PHASE III DUAL USE COMMERCIALIZATION: This technology should have a high commercial interest because it could also be used during humanitarian relief efforts in disaster areas and at remote construction sites.

REFERENCES:

1. Cost and Operational Effectiveness Analysis (COEA) for Gray Water Treatment to Support Force Provider and DEPMEDS, 26 March 1997, Prepared for US Army Tank Automotive Command under Contract Number DAAK70-92-D-0003, DO 0055.

2. Martel, C. J., Pelton, D. K. and Henry, K. S. (1999) Initial Evaluation of Geotextiles for Wastewater Filtration at Temporary Base Camps. To be presented at the Geosynthetics 99 Conference in Boston MA April, 1999.

KEYWORDS: Wastewater filtration, graywater treatment, sewage treatment.

ARMY99T008 TITLE: Development of a Multivalent, Oral Vaccine for Travelers

TECHNOLOGY AREAS: Biomedical

OBJECTIVE: To develop and evaluate a combination oral vaccine that provides active protection against the three main causes of travelers' diarrhea (TD): enterotoxigenic Escherichia coli (ETEC), Cammpylobacter jejuni, and Shigella species.

DESCRIPTION: TD is by far the most common illness in persons traveling from developed countries to lesser developed countries; nearly half of the 100 million such persons experience an episode of TD. During the Persian Gulf War, more than half of Coalition troops reported at least one episode of diarrhea within 3-4 months of deployment. More recently, rates of diarrhea ranging from 25% to 50% have been observed in ground forces participating in exercises in Thailand, Egypt, and South America. TD is also a frequent problem aboard U.S. Naval vessels following foreign port calls; more than a thousand man-hours were lost to TD among sailors and flight crews subsequent to a 3-day visit to Alexandria, Egypt, by the USS DWIGHT D EISENHOWER. Bacterial pathogens are the etiologic agents in at least 50% of all laboratory-confirmed cases of TD. The most common of these agents in enterotoxin-producing E. coli, or ETEC; Shigella and Campylocater are also important contributors to the overall burden of TD, and in some cases may be more significant than ETEC. The mix of diarrheal agents in dependent upon the geographic location, season and length-of-stay of the traveler. An effective vaccine against TD should be aimed at all three of the agents. In addition to the benefit to civilian travelers, such a vaccine would substantially reduce the risk for TD for deployed military personnel and thus contribute to force readiness. There are currently no vaccines available for TD; however, clinical challenge studies and examination of the natural history of diarrheal disease for TD; however, clinical challenge studies and examination of the natural history of diarrheal disease indicate that at least short-term protection is attainable. The Department of Defense has long maintained basic and applied research programs on each of the etiologic pathogens of TD; it is now feasible to consider combining the oral vaccine candidate products developed in these programs to produce a multivalent vaccine. A number of antigens, including lipopolysaccharide of Shigella, colonizqation factor antigens and the exterotoxin of ETEC, and flagellin of Campylobacter contribute to protective immunity. In addition, several new mucosal adjuvants may improve overall immunogenicity and protective efficacy when co-administered with monovalent or multivalent vaccines. It has recently been demonstrated that live-attenuated Shigella vaccines are highly immunogenic and protective in humans; the same approach to attenuation is now being adapted to all commonly-occurring Shigella serotypes. Preliminary studies in experimental animals show that these recombinant strains are also promising carriers of protective antigens for ETEC and Campylobacter.

PHASE I: Evaluate the potential of several targeted approaches as a basis for combined multi-agent TD vaccines. These approaches include vaccines comprised of: inactivated whole-cells, live-attenuated or live-recombinant Shigella carrier organisms, and subunit product candidates. Safety, immunogenicity, and the protective efficacy of selected vaccine prototypes will be assessed in experimental animals and in demonstration studies in volunteer human subjects.

PHASE II: Prepare clinical lots of the most promising combined vaccine candidates under Good Manufacturing Practices (GMP) procedures, conduct pre-clinical lot-release studies, and prepare documentation required by the U.S. Food and Drug Administration (FDA) in order to undertake human clinical trials under an Investigational New Drug (IND) application. Assess vaccine potency, safety, and immunogenicity in human volunteers. Evaluate mucosal adjuvants and various formulations with the experimental vaccines to optimize oral vaccine delivery schedule, dosage, and immunogenicity. Assess the protective efficacy of the most promising combined vaccine formulation in volunteer challenge studies.

PHASE III DUAL-USE COMMERCIALIZATION: Evaluate commercial markets for the potential of domestic, international, and DOD sales. Manufacture and assess consistency lots of vaccine produced under CMP, conduct expanded field safety and efficacy trials of the combined vaccine product in high-risk groups and if warranted, submit a product license Application (PLA) to the FDA.

KEY WORDS: Vaccine, multivalent, oral, traveler' diarrhea, Escherichia coli, ETEC, Shigella, Campylobacter, adjuvants, clinical trials, Department of Defense

ARMY99T009 TITLE: Development of Potential Therapeutic Compounds for Enhanced Membrane Permeability and Cholinergic Nerve Terminal Targeting

TECHNOLOGY AREAS: Biomedical

OBJECTIVE: Synthesis of Botulinum neurotoxin (BoNT) antagonists that can penetrate neuronal membranes and inhibit the catalytic light chain residing in the nerve terminal cytosol

DESCRIPTION: The seven BoNT serotypes are the most potent substances known to mankind. The neurotoxins act on cholinergic motorneurons to inhibit the release of acetylcholine leading to muscle weakness, paralysis and death. Exposure to BoNT generally arises from contact with contaminated food, septic wounds and colonization of infant digestive systems. An additional source for BoNT exposure is military conflict, since BoNT is a recognized BW agent that has been stockpiled by a number of hostile nations. At present, there are no approved post-exposure therapies for BoNT. With sudden conflicts, there may not be sufficient time to vaccinate large military and civilian populations to generate protective antibodies, thus there is an urgent need to develop effective post-exposure pharmacological therapies. Since the toxicity of BoNT stems from the zinc metalloprotease activity of the light chain, the most reasonable therapeutic approach is to develop inhibitors that are can interact with the active site zinc. Unfortunately, most currently-available zinc metalloprotease inhibitors are unable to penetrate biological membranes. This stems from their peptidomimetic nature and requirement for charged sulfonyl or sulphydryl groups for interacting with the active site zinc. It is therefore necessary to be able to modify these drugs to allow them to penetrate biological membranes, retain their efficacy as inhibitors and confine their actions specifically to cholinergic nerve terminals.

PHASE I: During this phase, membrane-permeable derivatives of available metalloprotease inhibitors shall be synthesized as prodrugs. The prodrugs should readily cross biological membranes and revert to their active form once inside the nerve terminal cytosol. The prodrug concept may be modelled on the acetoxymethylester analogs of the calcium indicator dye Fura-2, (Fura-2AM) which can readily penetrate biological membranes and become rapidly de-esterified to yield the active (charged) calcium indicator. Initial proof-of-concept may be performed with readily available metalloprotease inhibitors, such as captopril or phosphoramidon on appropriate isolated tissues or cell culture systems.

PHASE II: During this phase, bifunctional drugs will be developed for reduced systemic toxicity. To achieve this aim, it will be necessary to modify the drugs to also target cholinergic motorneurone terminals without compromising their ability to inhibit BoNT light chain or their ability to penetrate membranes. This may be accomplished by use of a linker that couples the active membrane-permeable drug to a component that recognizes unique markers on the cholinergic nerve ending such as the choline transporter or perhaps unique growth factor receptors. One approach is to develop a single low molecular weight drug (<500 kDa) that can bind selectively to the cholinergic nerve terminal, gain rapid entry to the cytosol and effectively inhibit the metalloprotease activity of BoNT. An alternative approach is to design more general molecules to achieve cholinergic membrane recognition that can be linked to available inhibitor molecules. These drugs will be tested on cholinergic spinal motoneurons and isolated rodent nerve-muscle preparations in conjunction with DoD investigators.

PHASE III DUAL USE APPLICATIONS: During this phase, a small number of drugs that exhibit optimal metalloprotease inhibitor properties, the best cholinergic targeting systems and the most efficient membrane penetration systems will be tested on cells, isolated tissues and in vivo. In addition to botulism, other potential benefits of this technology include development of other classes of drugs that recognize the cholinergic nerve terminal. This would be highly desirable for delivering therapeutic substances in a number of disease states such as the Eaton-Lambert Myasthenic Syndrome where acetylcholine release is insufficient to maintain normal muscle activity. The technology would also facilitate development of targeted drugs to other nerve endings and may be beneficial in Alzheimer's dementia or Parkinson's disease. The advantage of this system over those that use non-toxic genetically altered BoNT as the delivery vehicle is that it would not require endosomal partitioning of drugs and use of large (potentially unstable) proteins for recognition and delivery.

KEY WORDS: Botulinum toxin, metalloprotease inhibitors, cholinergic